

Analysis of Fat-Soluble Vitamins using the Kinetex® Core-Shell Biphenyl LC Column

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Overview

Fat-soluble vitamins, such as vitamin A (Retinol) and E (α - and γ -Tocopherol), are carried to the body's fat tissues and stored. Being able to successfully separate these fat-soluble vitamins is a useful tool for analytical and clinical labs. In this application note, we present an LC-MS/MS method to separate fat-soluble vitamins, and their different analogues, by utilizing the Kinetex 2.6 μ m Biphenyl LC column.

In this method, a panel of fat-soluble vitamins was separated using a core-shell particle with a biphenyl surface chemistry. The Kinetex Biphenyl offers the high-performance benefits of a core-shell particle with enhanced retention, higher sensitivity, and overall better results for the analyte panel, particularly for the two analogues of Vitamin E (α - and γ -Tocopherol).

The optimal signal sensitivity under the prescribed LC mobile phase condition was achieved while the mass spectrometry method used a dual polarity under APCI mode in order to view the vitamin ions with their different polarity. The Kinetex Biphenyl column is the only column that allows enough time to switch the polarity without losing any sample, but also provides a fast run time. Together, this method offers users a unique option when developing a method to separate fat-soluble vitamins.

LC-MS/MS Conditions

Column: Kinetex 2.6 µm Biphenyl

Dimension: 50 x 3.0 mm **Part No.:** 00B-4622-Y0 **Mobile Phase:** A: Water

B: Acetonitrile/Isopropyl alcohol (1:1)

| Gradient: | Time (min) | %B | Flow Rate (mL/min) |
|------------------|------------|----|--------------------|
| | 0 | 65 | 0.6 |
| | 2 | 95 | 0.6 |
| | 2.01 | 95 | 0.5 |
| | 3.5 | 95 | 0.5 |
| | 3.51 | 65 | 0.6 |
| | 5 | 65 | 0.6 |

Injection Volume: $10 \mu L$ Temperature: $25 \, ^{\circ}C$

LC System: Agilent® 1260 Infinity

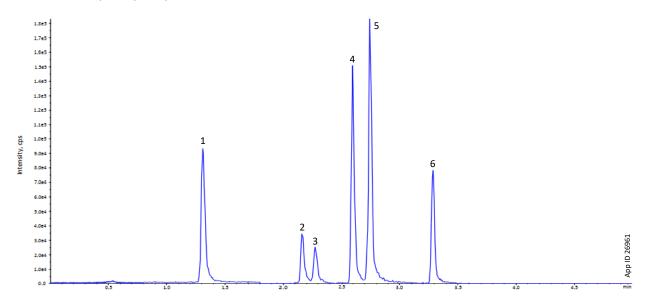
Detection: LC-MS/MS

Detector: 6500+ QTRAP® (SCIEX®), APCI (Dual Polarity)

MRM Transitions

| Peak No. | Analyte | Q1 (m/z) | Q3 (m/z) | | |
|----------|--------------------------|-------------|-------------|--|--|
| 1 | Vitamin A (Retinol) | 269.2 | 93 | | |
| 2 | Vitamin E (γ-Tocopherol) | 415.3 | 149.22 | | |
| 3 | Vitamin E (α-Tocopherol) | 429.4 | 163.2 | | |
| 4 | Vitamin K2 (MK-4) | 445.4 | 187.1 | | |
| 5 | Vitamin K1 | 451.3 | 187.2 | | |
| 6 | Vitamin K2 (MK-7) | 649.5 | 187 | | |

Kinetex® 2.6 μm Biphenyl



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